(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
6 January 2005 (06.01.2005)

PCT

(10) International Publication Number
WO 2005/000258 A1

(51) International Patent Classification[7]: A61K 7/06

(21) International Application Number:
PCT/KR2004/001572

(22) International Filing Date: 28 June 2004 (28.06.2004)

(25) Filing Language: Korean

(26) Publication Language: English

(30) Priority Data:

10-2003-0042703	27 June 2003 (27.06.2003)	KR
10-2003-0087283	3 December 2003 (03.12.2003)	KR
10-2004-0013452	27 February 2004 (27.02.2004)	KR
10-2004-0044544	16 June 2004 (16.06.2004)	KR
10-2004-0044542	16 June 2004 (16.06.2004)	KR

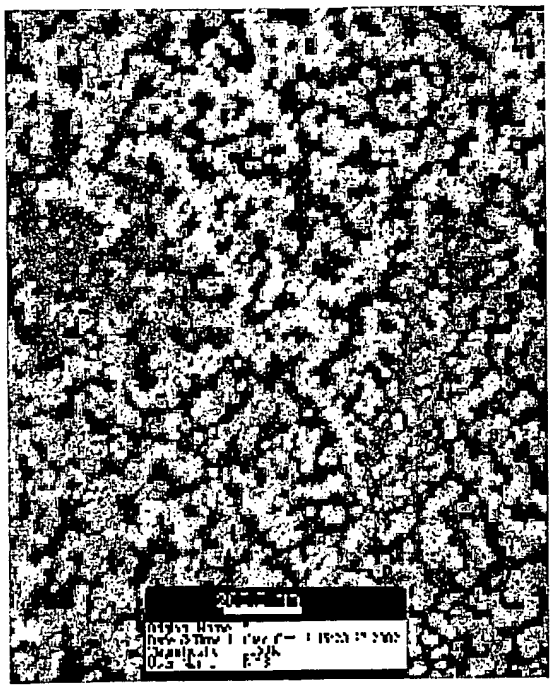
(71) Applicant *(for all designated States except US)*: **AMOREPACIFIC CORPORATION** [KR/KR]; 181, Hangang-ro 2-ga, Yongsan-gu, Seoul 140-777 (KR).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **NAM, Yoon Sung** [KR/KR]; No. 901-204 Jugong Apartment, Sinjung Village, Poongduckchon-ri, Suji-eup, Yongin-si, Kyunggi-do 449-846 (KR). **SHIM, Jong Won** [KR/KR]; No. 217-202 Jugong-2-danji Apartment, Banpo-dong, Seocho-gu, Seoul 137-766 (KR). **KANG, Byung Young** [KR/KR]; No. 308-1503 Mido-1-cha Apartment, Banpo-4-dong, Seocho-gu, Seoul 137-769 (KR). **HWANG, Jae Sung** [KR/KR]; No. 311-1503 Ssangyong Apartment 621 Sangha-ri, Gusung-myun, Yongin-si, Kyunggi-do 449-914 (KR). **KIM, Jun Oh** [KR/KR]; 514-1 Yatap-dong, Bundang-gu, Sungnam-si, Kyunggi-do 463-070 (KR). **CHANG, Ih Seop** [KR/KR]; No. 102-1104 Dongbo Apartment 703 Poongduckchon-ri, Suji-eup, Yongin-si, Kyunggi-do 449-846 (KR). **PARK, Won Seok** [KR/KR]; No. 401 elitevil 749-12, Yoksam-dong Kangnam-gu, Seoul 135-080 (KR). **KANG, Hyung Seok** [KR/KR]; No. 302 Ihwavilla, 933-4, Daechi-4-dong, Kangnam-gu, Seoul 135-844 (KR). **LEE, Byung Seok** [KR/KR]; No. 1-121 Pyungchang Apartment, Pyungchang-dong, Jongro-gu, Seoul 110-012 (KR). **CHO, Won Hee** [KR/KR]; No. 71-1305 Hyundai Apartment, Apgujung-dong, Kangnam-gu, Seoul 135-788 (KR). **SUNG, Dae Seok**

[Continued on next page]

(54) Title: SELF-ASSEMBLED POLYMERIC NANOPARTICLES CONTAINING PHYSIOLOGICALLY ACTIVE INGREDIENTS AND EXTERNAL APPLICATION CONTAINING THE NANOPARTICLES



(57) Abstract: The present invention relates to self-assembled polymeric nanoparticles containing physiologically active ingredients and to an external application containing the nanoparticles, in particular, relates to a self-assembled polymeric nanoparticles having amphiphilic polymeri, which comprises polycaprolactone as a hydrophobic block and polyethyleneglycol as a hydrophilic block to solubilize and entrap physiologically active ingredients in an aqueous solution, and to an external application for skin containing the particles.

WO 2005/000258 A1

[KR/KR]; No. 4-817 Miju Apartment, Chungryangri-1-dong, Dongdaemoon-gu, Seoul 130-781 (KR). **KIM, Dae Kwon** [KR/KR]; No. 206 Campustel, 1566-16, Bongchon-4-dong, Kwanak-gu, Seoul 151-054 (KR). **LEE, Chang Hoon** [KR/KR]; No. 202-602 Hyundai-hometown, Gaebong-dong, Guro-gu, Seoul 152-090 (KR). **HAN, Sang Hoon** [KR/KR]; No. 2-203 Chunrok Apartment, 276-3, Yooljun-dong Changan-gu, Suwon-si, Kyunggi-do 440-320 (KR). **SIM, Young Chul** [KR/KR]; No. 123-1402 Samik Apartment Parktown, Sunae-dong, Bundang-gu, Sungnam-si, Kyunggi-do 463-020 (KR). **YEOM, Myeong Hoon** [KR/KR]; No. 104-1101 Byeoksan Apartment, Jookjun-2-dong, Suji-eup, Yongin-si, Kyunggi-do 449-755 (KR). **LEE, Sung Il** [KR/KR]; No. IB-1512 Kolontripolice, Kumgok-dong, Bundang-gu, Sungnam-si, Kyunggi-do 463-480 (KR). **KIM, Hyoung June** [KR/KR]; No. 610-1004 Byeoksan Apartment, Yatap-dong, Bundang-gu, Sungnam-si, Kyunggi-do 463-927 (KR). **YANG, Hye Jin** [KR/KR]; No. 208-304 Wooman-Jugong Apartment, Wooman-2-dong, Paldal-gu, Suwon-si, Kyunggi-do 442-753 (KR).

(74) **Agent: YOON, Dong Yol**; 9th Fl., Yosam Bldg., 648-23, Yoksam-dong, Kangnam-gu, Seoul 135-748 (KR).

(81) **Designated States** *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) **Designated States** *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

— *with international search report*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

Self-assembled polymeric nanoparticles containing physiologically active ingredients and external application containing the nanoparticles

FIELD OF THE INVENTION

The present invention relates to self-assembled polymeric nanoparticles containing physiologically active ingredients and to an external application containing the nanoparticles. In particular, the present invention provides a self-assembled polymeric nanoparticle having amphiphilic polymer, which comprises polycaprolactone as a hydrophobic block and polyethyleneglycol as a hydrophilic block to solubilize and to entrap physiologically active ingredients in an aqueous solution, and provides an external application for skin containing the particles.

BACKGROUND OF THE INVENTION

Recently, methods for preparing nanometer- or micrometer-sized emulsion particles comprising medical substances, lipid, glycerol, water and phospholipid or nonionic surfactants are reported (USP 5,338,761), and other method having phospholipid with electronic charge as an emulsifier is also reported (USP 6,120,751). For Example, nano-emulsion is prepared by treating semi-formulated emulsion obtained by using surfactants having specific hrdrophilc-hydrophobic ratio with a high-pressure emulsifier (emulsifying machine), and liposome is a spherical or indeterminate formed particle having multi-layered membranes

made of lipid from vegetables or animals in which various materials are captured. The two formulations are widely used in cosmetics. In addition, a method for preparing nanosized emulsion using microemulsions obtained when 3 phases of emulsifier, oil and water get reached to a certain concentration (USP 5,152,923, WO 91/06,286 and WO 91/06,287).

However, in the above conventional art, because membranes of emulsion particles are in a state of dynamic equilibrium with outer phase, the ingredients in the emulsion contact with water, which causes oxidation or decomposition of the ingredient making the particles degenerated. In addition, because the membranes of the emulsion particles are chemically or physically weak and unstable, the membranes are easily broken by organic or inorganic pollutants, and a long-terms of storage is nearly impossible. Therefore, the nanosized particles prepared by using low-molecular-weight emulsifier are not sufficient to be used for unstable ingredients, and formulation thereof is difficult. In addition, there is a problem that a large amount of surfactants should be used to contain high concentration of effective ingredients, which causes skin irritation.

However, using nanoemulsion particle has such effects that the effective ingredient therein are simultaneously released from the particle when applied to the skin because the membrane of the particle breaks on the skin or breaks in the skin after absorbed thereto, and that molecular design of membrane compound is possible which enables to decrease contact with outer phase. For Example, a method using cochleate to minimize the contact of the inner materials with outer phase and to increase the release of ingredients is reported (USP 4,663,161). So, when low molecular weight materials are used, a technique to improve chemical

and physical stability of the emulsion particle and chemical stability of the inner physiological ingredients is required. Particularly, nano-techniques that enables the physiological ingredients captured into the particle to be released effectively from formulation are required.

In order to overcome the weak points of the emulsion particles made from low molecular weight materials, a method using polymers instead of lipid as for hydrophobic core is reported, wherein polymers are dissolved in a solvent using excessive amount of surfactants and dispersed to nanometer sizes then solidified by distilling solvent (Colloids and Surface A, 210(2002), 95-104).

Because the nanoparticles are very small and show colloidal instability, various surfactants or stabilizers are used in the conventional method and such procedures as high-pressure emulsification consuming a lot of energy should be applied to make nanosized particles. In addition, the conventional methods have the problems of Ostwald ripening, precipitation or flocculation due to colloidal instability, and the colloidal instability becomes serious when the amount of solid components are increased, therefore the particles prepared by the conventional method can not contain a lot of effective ingredients (21th Proceedings of IFSCC International Congress, 2000(2000), 442-458).

In order to overcome the problems of the conventional methods, various novel methods to capture the effective components stably have been studied, which is very important for external application, especially, in the field of cosmetics or pharmaceuticals. In particular, in the field of growing hairs, a novel liposome is being researched to deliver and supply effective materials to the hair bulb stably (Follicular liposomal delivery systems, J. Liposome Res., 2002, 12:

143-8), and nanosized carriers are also studied widely.

SUMMARY OF THE INVENTION

The present invention relates to self-assembled polymeric nanoparticles containing physiologically active ingredients and to an external application containing the nanoparticles, in particular, to a self-assembled polymeric nanoparticle having amphiphilic polymeri, which comprises polycaprolactone as a hydrophobic block and polyethyleneglycol as a hydrophilic block to solubilize and to entrap physiologically active ingredients in an aqueous solution, and to an external application for skin containing the particles.

The self-assembled polymeric nanoparticles of the present invention are very useful to formulate and stabilize water-insoluble physiological components, this is because the polymeric nanoparticle of the present invention has a property of capturing insoluble components due to its self-assemble characteristic.

Physiological components that would be captured in the self-assembled polymeric nanoparticles of the present invention may comprise ginsenoside, co-enzyme Q10, active components for growing hairs such as finasteride and cyclosporin, but not restricted thereto.

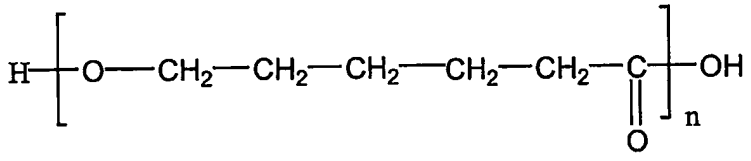
DETAILED DESCRIPTION OF THE INVENTION

The amphiphilic polymer having self-assembling characteristic and being applied to prepare the nanoparticle of the present invention is, preferably, a

copolymer of hydrophobic biodegradable polycaprolactone (PCL, Formula 1; component "A") and hydrophilic biodegradable polyethyleneglycol (PEG, Formula 2; component "B"). Even though A-B type double block copolymer or A-B-A or B-A-B type triple block copolymer is most preferable, multiple block type or graft type copolymer is also acceptable and the type of copolymer is not restricted.

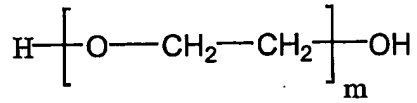
Preferably, the hydrophobic polymer may be PCL with a molecular weight of 500 to 100,000 daltons, more preferably, 1000 to 25,000 daltons. The hydrophilic polymer may be PEG with a molecular weight of 500 to 100,000 daltons, more preferably, 1000 to 25,000 daltons. The ratio of the PCL and the PEG is preferably 1:9 to 9:1 by weight, more preferably 3:7 to 7:3, and most preferably, the ratio of the PCL and the PEG is 6:4 by weight.

[Formula 1]



wherein, n is an integer of 2 or more than 2.

[Formula 2]



wherein, m is an integer of 2 or more than 2.

The bonding of polycaprolactone and polyethyleneglycol of the present

invention is preferably covalent bonding such as ester bonding, anhydride bonding, carbamate bonding, carbonate bonding, amine bonding, amide bonding, secondary amine bonding, urethane bonding, phosphodiester bonding or hydrazone bonding.

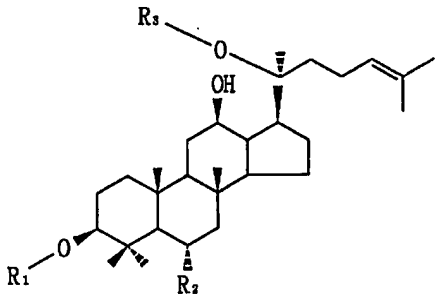
The physiological components captured and contained in the nanoparticle of the present invention may be such materials that can be solubilized in the polymer, especially, water-insoluble components that could not be formulated by the conventional method, for Example, ginsenosides, coenzyme Q10, hair growing components, but not restricted thereto.

For Example, *Rheum undulatum*, genistein, hesperetin, hesperidine, catechin, isoflavone, danazol, haloperidol, furosemid, isosorbide dinitrate, chloramfenicol, sulfamethoxazole, caffeine, cimethidine, diclofenac Na, coenzyme Q10, vitamin E and its derivatives, vitamin A and its derivatives, provitamin D_3 and its derivatives, ursolic acid, oleanolic acid, rosmarinic acid, 18 beta-glycyrrhetinic acid, glabridin, aleuritic acid, polyphenol, esculin, (-) epigallocatechin gallate, turmeric acid, ginsenosides, tetra hydrocurcuminoids, centella asiatica, beta carotene, asiaticoside, farnesol, beta-sitosterol, linoleic acid, gamma linolenic acid, resveratrol, vineatrol, ginkgo biloba, triclosan, minoxidil, natural oil, ceramide, sphingosine, extracts of Thujae occidentalis, extracts of Polygoni multiflori Radix, extracts of Glycyrrhiza uralensis, extracts of Coix lachryma-jobi var. ma-yuen and finasteride may be comprised therein.

In particular, ginseng saponins, especially ginsenosides represented by Formula 3, for Example, ginsenoside Rh1, Rh2, F1 (Formula 4a) and compound K (Formula 4b) having a structure that a glucose is bonded to ginseng aglycon,

and 20-O-[-L-arabinopyranosyl(1->6)-D-glucopyranosyl]-20(S)-protopanaxadiol having a structure that two glucoses are bonded to ginseng aglycon are useful for restricting proliferation of cancer cells or tumor cells, improving the activity of anticancer agents.

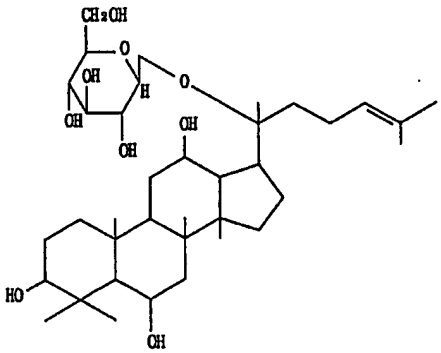
[Formula 3]



In the Formula 3, R1, R3 is glucose or H; R2 is golucose, H or OH; and at least one of R1, R2 and R3 is glucose. Ginsenoside is a kind of ginseng saponin, and any type of ginsenoside can be applied in the present invention. That is, crude type extracted from ginseng or bio-transformed type can be applied.

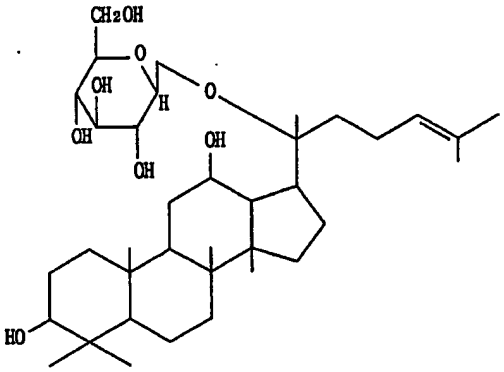
Ginsenosides represented by Formula 3 are preferable.

[Formula 4a]



Ginsenoside F1

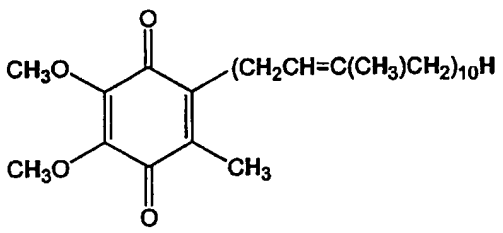
[Formula 4b]



Compound K

In addition, coenzyme Q10 represented by following Formula 5 may be usefully contained into the self-assembled polymeric nanoparticles of the presenti invention.

[Formula 5]



In addition, the self-assembled polymeric nanoparticles of the present invention may be applied to contain and carry the active components for growing and sprouting hairs that are effective but difficult to be formulated. Examples of such components for growing and sprouting hairs comprises finasteride, minoxidil, extracts of Thujae occidentalis, extracts of polygoni multiflori Radix, extracts of Glycyrrhiza uralensis, extracts of Coix lachryma-jobi var. ma-yuen, isoflavone, genistein, hesperetin, hesperidine, catechin, vitamin E and its derivatives, vitamin A its derivatives, provitamin D3 and its derivatives, ursolic acid, oleanolic acid, rosmarinic acid, 18-beta-glycyrrhetinic acid, farnesol, beta-sitosterol, linoleic acid, gamma linolenic acid, resveratrol, ceramide, Sphingosine, or the like.

In particular, cyclosporin, an important immunosuppressive agent that is administered to a patient of organ transplantation by orally and used to treat psoriasis, was applied to the apopecia areata, and it is reported that cyclosporin

has activities of growing hairs in the animal experiments. Finasteride, an active component for growing and sprouting hairs, is a specific inhibitor to the second type 5α -reductase, and when it is orally administered, it prevents transformation of testosterone to dihydrotestosterone, so it is used for treating prostatitis or apopecia areata. The above insoluble physiological components can be purchased or prepared by one skilled in the art to be applied to the present invention. And, vegetable extracts are easily obtained or prepared.

A method for preparing self-assembled polymeric nanoparticle containing physiologically active components comprises the following steps of:

(a) Preparing amphiphilic polymer comprising polycaprolactone as a hydrophobic block and polyethyleneglycol as a hydrophilic block to form block copolymer;

(b) Dissolving the amphiphilic polymer and physiologically active components in an organic solvent and stirring to prepare solution mixture; and

(c) Pouring the solution mixture prepared through (a) and (b) a water solution to obtain nanoparticles; and

(d) Removing organic solvent.

The amount of physiologically active components contained in the nanoparticles may be controlled according to its use and object, and preferably, 1 to 50wt% to the total weight of the nanoparticles, more preferably, 20 to 50wt%. When the amount of physiologically active components is more than 50%, the physiologically active components are not effectively entrapped and outflow

from the nanoparticles, which causes cohesion and so causes discoloration or change of odor.

The mean size of the nanoparticles is preferably 1 to 1,000nm, more preferably, 10 to 500nm.

Methods for preparing self-assembled polymeric nanoparticle containing physiologically active components using the PCL-PEG copolymer of the present invention in an aqueous solution comprises a method dispersing the PCL-PEG copolymer and applying supersonic waves; a method dispersing or dissolving the copolymer in an organic solution then removing the organic solvent by extracting with excessive water or by distilling away; a method dispersing or dissolving the copolymer in an organic solution and stirring severely with homogenizer or high pressure emulsifier then distilling away the solvent; a method dispersing or dissolving the copolymer in an organic solution then dialyzing with excessive water; a method dispersing or dissolving the copolymer in an organic solution then adding water slowly; or the like.

Organic solvent for biodegradable PCL-PEG copolymer to prepare polymeric nanoparticle of the present invention in an aqueous solution is at least one selected from the group consisting of acetone, dimethylsulfoxide, dimethylformamide, N-methylpyrolidone, dioxane, teterhydrofuran, ethylacetate, acetonitryl, methylethylketone, methylenechloride, chloroform, methanol, ethanol, ethylehter, diethylether, hexane, petroleum ether, or mixture thereof.

In the nanoparticle of the present invention, physiologically active components are captured in a hydrophobic core of the self-assembled polymeric nanoparticle and hydrophilic polymer chain is arranged on the surface of the

nanoparticle, which makes the nanoparticle stably disperse in aqueous phase. The nanoparticle dispersed in the aqueous phase has nanometer particle size and colloidal stability is very high. When this nanoparticle is applied to skin external application composition, the composition is stable because the physiologically active components do not directly contact with formulation components or with skin, and easily formulated as cream, lotion, cosmetic water, or the like. The skin external application prepared above has the effects of the physiologically active components to improve skin, and shows improved absorption property into the skin, scalp or hair bulb. In addition, the self-assembled amphiphilic polymer of the present invention is degraded in a body, and so very safe.

The formulation of the external application of the present invention is restricted and may be any formulation of hair tonic, scalp-treatment, hair cream, ointment, soft water, skin softener, nutrition water, eye cream, nutrition cream, massage cream, cleansing cream, cleansing foam, cleansing water, powder, essence, pack, body lotion, body cream, body oil, body essence, make-up base, foundation, hair dye, shampoo, rinse, body cleaner, tooth paste, oral cleaner, lotion, gel, patch or spray. In addition, any components that are soluble to the solvent used for the preparation of the application may be selected and added by one skilled in the art according to its use and object.

BRIEF DESCRIPTION OF THE DRAWINGS

Fig. 1 is a feature of self-assembled polymeric nanoparticle containing Q10 coenzyme taken by transmission electron microscope.

Fig. 2 is a feature of self-assembled polymeric nanoparticle containing the extracts of Thujae occidentalis taken by transmission electron microscope.

Fig. 3 is a feature of self-assembled polymeric nanoparticle containing ginsenoside taken by transmission electron microscope.

Fig. 4 is a feature of self-assembled polymeric nanoparticle containing minoxidil being absorbed into the hair bulb of the hairy Guinea.

Fig. 5 is a feature of self-assembled polymeric nanoparticle containing minoxidil being absorbed into the hamster flank organ.

PREFERRED EMBODIMENT OF THE INVENTION

Hereinafter, the present invention is described with reference to Examples and Experimental Examples. However, the scope of the invention is not restricted by the Examples.

[Reference Example 1] Preparation of ginsenoside (purified ginseng saponin)

2kg of Red ginseng (KT&G Corporation) was added into 4ℓ of methanol containing water, and refluxed 3 times for extraction then deposited for 6 days at 15℃. Residues and remainders were separated by filtration and centrifugation, then the remainders were concentrated under reduced pressure to obtain extract. The extract was suspended into the water and re-extracted with 1ℓ of ether 5 times to remove pigments, then water layer thereof was extracted with 500ml of 1-butanol 3 times. The above-obtained 1-butanol layer was treated with 5% of KOH and washed with distilled water then concentrated under reduced pressure

to obtain 1-butanol extract. The extract (1-butanol extract) was dissolved in a small amount of methanol, and a large amount of ethylacetate was added thereto to obtain precipitation. The precipitation was dried to obtain 100g of purified ginseng saponin extract (yield: 5%).

[Reference Example 2] Preparation of gensenoside with enzyme hydrolysis

10g of purified ginseng saponin obtained in the Reference Example 1 was dissolved in 100ml of citrate buffer solution (pH 5.5), and 1g of naringinase obtained from *penicillium sp.* and 1g of pectinase obtained form *Aspergillus sp.* was added thereto and reacted for 48 hours while stirring at 40℃ in water bath. Thin layer chromatograph was performed to check whether the substrate was consumed, and after completeste consumption of substrate, reaction mixture was heated for 10 minutes to terminate the reaction. Then the reaction mixture was extracted 3 times with same amount of ether then filtered and concentrated to obtain 1,050mg of ginseng saponin mixture treated with enzyme comprising 440mg of Compound K, 150mg of ginsenoside F1 and other ginsenosides with 1~4 of glucose (yield: 10.5%).

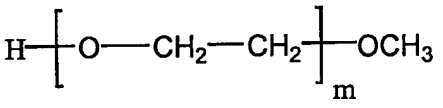
[Preparation Examples 1~9] Preparation of PCL-PEG block copolymer

The method for preparation of PCL-PEG block copolymer described in this Example is only for reference, the copolymer of the present invention is not restricted thereto.

The PCL-PEG block copolymer of the present invention was prepared by open-ring polymerization of polycaprolactone monomers.

Specific amount of methoxy PEG [hereinafger, we call "mPEG" (Foumula 6)] and catalyst of $Sn(Oct)_2$ (Sigma, St. Louis, MO, U.S.A.) according to Table 1 was added into a glass flask containing hexamethyldisilazine silanizated by the reaction with hydroxy group, and polycaprolactone monomers was also added and homogeneously mixed. Said mPEG (Fluka Chemie GmbH, Buchs, Swiss) is a PEC, one terminal of which is substituted with methoxy to prevent reaction and only the other side of terminal hydroxy group can make polymerization reaction.

[Formula 6]



The flask containing the mixture was connected with vacuum line, and water was removed under vacuum and sealed up, then stayed at 120℃ for polymerization. After 24 hours later, polymerized polymer was dissolved in methylene chloride, and re-crystallized with excessive methanol to obtain pure PCL-PEG copolymer.

Molecular weight of the above-obtained PCL-PEG was measured with gel permeation chromatography (hereinafter, we call "GPC"). The GPC was Agilent 110 series (Agilent Technologies, Palo Alto, CA, U.S.A.), polymer was detected with Refractive Index (RI) detector, columns were three PLgel columns (300 x 7.5 mm, pore size = 10^3, 10^4 and 10^5 Å), flow rate was 1.0ml/min, and mobile phase was tetrahydrofuran (THF)

[Table 1] Preparation of PCL-PEG block copolymer

	PCL monomer (g)	mPEG (g)	M. W. of mPEG (dalton)	$Sn(Oct)_2$ (g)	M. W. of PCL-PEG polymerized (dalton)
P. E. 1	33	66	5,000	0.5	6,900
P. E. 2	50	50	5,000	0.5	8,500
P. E. 3	60	40	5,000	0.5	11,000
P. E. 4	66	33	5,000	0.5	13,500
P. E. 5	75	25	5,000	0.5	18,700
P. E. 6	80	20	5,000	0.5	24,300
P. E. 7	50	50	3,500	0.5	5,400
P. E. 8	50	50	2,000	0.5	3,200
P. E. 9	50	50	1,000	0.5	1,700

P.E.: Preparation Example, M.W.: Molecular weight

[Preparation Examples 10~11] Preparation of polycaprolactone-co-polyethylene-glycol block copolymer

10g of monomethoxypolyethyleneglycol (M.W. 5,000) of which one terminal is hydroxy group and 10g of caprolactone monomer were added to a 50mL dried flask, and 0.05g of $Sn(Oct)_2$ as a catalyst was added thereto. A magnetic bar coated with Teflon was dropped to the flask, and the mixture underwent vacuum treatment for 30 minutes, then the flask was sealed tightly. The above sealed flask was put in 150℃ oil bath and performed polymerization for 6 hours.

The polymerization product was hard solid state, so the polymerization product was dissolved with 20ml of methylenechloride completely and precipitated with excessive ethylether. This procedure was repeated 3 times to remove not-reacted monomers and oligomers. The product of precipitation was vacuum dried at room temperature for 12 hours, and finally 17.3g of polycaprolactone-block-polyethyleneglycol block copolymer was obtained.

It was verified by ^{1}H NMR analysis that polycaprolactone at a terminal of monoethoxypolyehtyleneglycol was ring-opened to perform co-polymerization. By the integration of peaks indicating monoethoxypolyehtyleneglycol and polycaprolactone, it was found that number average molecular weight (Mn) was 9,200.

100g of polyethyleneglycol having primary amine at a terminal [O,O'-Bis-(aminopropyl) polypropylene glycol-block-polyethylene glycol-block-polypropylene glycol; weight average molecular weight (Mw) 1,900] and 20g of acetone was added to 500ml reaction flask, and heated to 90℃ to dissolve. The above dissolved solution was heated to 100℃, and 100g of polycaprolactone (Mw; 80,000) was added, then stirred for 1 hour with 100rpm. The prepared homogeneous solution was stirred for 5 hours with 300rpm, then cooled to room temperature. After completing reaction, the polymer product was dispersed and stirred in distilled water to purify and to remove not-reacted polyethyleneglycol, which was repeated 3 times, and finally obtained 188g of block copolymer of polyethyleneglycol and polycaprolactone.

[Preparation Examples 12~13] Preparation of block copolymer of polyethylene-glycol and poly-D,L-lactic acid-co-glycolic acid

5g of monomethoxypolyethyleneglycol (M.W. 5,000) of which one terminal is hydroxy group, 7g of D,L-lactic acid and 3g of glycolic acid were added to a 20mL dried flask, and 0.025g of $Sn(Oct)_2$ as a catalyst was added thereto. A magnetic bar coated with Teflon was dropped to the flask, and the mixture underwent vacuum treatment for 30 minutes, then the flask was sealed

tightly. The above sealed flask was put in 130℃ oil bath and performed polymerization for 6 hours.

The polymerization product was hard solid state, so the polymerization product was dissolved with 20ml of methylenechloride completely and precipitated with excessive ethylether. This procedure was repeated 3 times to remove not-reacted monomers and oligomers. The product of precipitation was vacuum dried at room temperature for 12 hours, and finally 12.7g of block copolymer of polyethyleneglycol and poly-D,L-lactic acid-co-glycolic acid.

It was verified by ^{1}H NMR analysis that D,L-lactic acid and glycolic acid at the terminal of monoethoxypolyehtyleneglycol was ring-opened to perform co-polymerization. By the gel permeation chromatography (GPC), it was found that number average molecular weight (Mn) was 12,500.

2g of polyethyleneglycol having primary amine at a terminal [O,O'-Bis-(aminopropyl) polypropylene glycol-block-polyethylene glycol-block-polypropylene glycol; weight average molecular weight (Mw) 900] was added to 50ml reaction flask, and heated to 90℃ to dissolve. The above dissolved solution was heated to 100℃, and 20g of poly-D,L-lactic acid-co-glycolic acid (RG502, Boehringer Ingelheim; Mw 11,000) was added, then stirred for 1 hour with 100rpm. The prepared homogeneous solution was stirred for 3 hours with 300rpm, then cooled to room temperature. After completing reaction, the polymer product was dispersed and stirred in distilled water to purify and to remove not-reacted polyethyleneglycol, which was repeated 3 times, and finally obtained 20.4g of block copolymer of polyethyleneglycol and poly-D,L-lactic acid-co-glycolic acid.

[Preparation Example 14] Preparation of poly-D,L-lactic acid-co-polyethyleneglycol copolymer

5g of monomethoxypolyethyleneglycol (M.W. 5,000) of which one terminal is hydroxy group and 5g of D,L-lactic acid were added to a 20ml dried flask, and 0.025g of $Sn(Oct)_2$ as a catalyst was added thereto. A magnetic bar coated with Teflon was dropped to the flask, and the mixture underwent vacuum treatment for 30 minutes, then the flask was sealed tightly. The above sealed flask was put in 130℃ oil bath and performed polymerization for 6 hours.

The polymerization product was hard solid state, so the polymerization product was dissolved with 20ml of methylenechloride completely and precipitated with excessive ethylether. This procedure was repeated 3 times to remove not-reacted monomers and oligomers. The product of precipitation was vacuum dried at room temperature for 12 hours, and finally obtained 13.1g of poly-D,L-lactic acid-block-polyethyleneglycol block copolymer.

It was verified by ^1H NMR analysis that D,L-lactic acid at the terminal of monoethoxypolyehtyleneglycol was ring-opened to perform co-polymerization. By the gel permeation chromatography (GPC), it was found that number average molecular weight (Mn) was 13,700.

[Preparation Example 15] Preparation graft block copolymer of polyethyleneimine and polycaprolactone

2g of graft polyethyleneimine having primary amine at a terminal (Mw; 900) was added to 50ml reaction flask, and heated to 90℃ to dissolve. The

above dissolved solution was heated to 100℃, and 20g of polycaprolactone (Mw; 80,000) was added, then stirred for 1 hour with 100rpm. The prepared homogeneous solution was stirred for 5 hours and cooled to room temperature. After completing reaction, the polymer product was dispersed and stirred in distilled water to purify and to remove not-reacted polyethyleneglycol, which was repeated 3 times, and finally obtained 21.2g of block copolymer of polyethyleneimine and polycaprolactone.

[Preparation Example 16] Preparation linear block copolymer of polyethyleneimine and polycaprolactone

2g of linear polyethyleneimine having primary amine at a terminal (Mw; 900) was added to 50ml reaction flask, and heated to 90℃ to dissolve. The above dissolved solution was heated to 100℃, and 20g of polycaprolactone (Mw; 80,000) was added, then stirred for 1 hour with 100rpm. The prepared homogeneous solution was stirred for 5 hours and cooled to room temperature. After completing reaction, the polymer product was dispersed and stirred in distilled water to purify and to remove not-reacted polyethyleneglycol, which was repeated 3 times, and finally obtained 19.1g of block copolymer of polyethyleneimine and polycaprolactone.

[Examples 1-20] Preparation of polymeric nanoparticles containing ginsenoside prepared by using polycaprolactone-co-polyethyleneglycol block copolymer

Polycaprolactone-co-polyethyleneglycol block copolymer (Mw = 10,000 dalton, polycaprolactone : polyethyleneglycol = 1:1 by weight) and ginsenoside

were dissolved in 50ml of organic solvent, then the mixture solution was poured into 50ml of aqueous solution to induce self-assembling to form nanoparticles. The organic solvent was removed by distillation or dialysis to obtain aqueous solution of nanoparticles containing ginsenoside. The ginsenosides used in the Examples were those of Reference Examples 1 and 2, which were obtained by treating the saponin extracted from *Panax ginseng* C. A. Meyer (*Araliaceae*) with enzyme.

[Table 2] Preparation condition of the nanoparticles containing ginsenoside

	Amphiphilic polymer and amount used	Ginsenoside	Organic solvent	Removal of organic solvent
Example 1	P. E. 1, 1.5g	0.5g	Ethanol	Distillation
Example 2	P.E. 1, 10g	1.2g	Ethanol	Distillation
Example 3	P.E. 1, 25g	2.5g	Ethanol	Distillation
Example 4	P.E. 2, 1.5g	0.5g	Ethanol	Distillation
Example 5	P.E. 2, 10g	1.2g	Ethanol	Distillation
Example 6	P.E. 2, 25g	2.5g	Ethanol	Distillation
Example 7	P.E. 3, 1.5g	0.5g	Ethanol	Distillation
Example 8	P.E. 3, 10g	1.2g	Ethanol	Distillation
Example 9	P.E. 3, 25g	2.5g	Ethanol	Distillation
Example 10	P.E. 4, 8g	1.2g	Ethanol	Distillation
Example 11	P.E. 4, 8g	1.2g	Dimethylsulfoxide	Dialysis
Example 12	P.E. 4, 8g	1.2g	Dimethylformamide	Dialysis
Example 13	P.E. 4, 8g	1.2g	Aceconitryl	Dialysis
Example 14	P.E. 4, 8g	1.2g	Tetrahydrofuran	Dialysis
Example 15	P.E. 4, 8g	1.2g	Acetone	Dialysis
Example 16	P.E. 5, 8g	1.2g	Ethanol	Distillation
Example 17	P.E. 6, 8g	1.2g	Ethanol	Distillation
Example 18	P.E. 7, 8g	1.2g	Ethanol	Distillation
Example 19	P.E. 8, 8g	1.2g	Ethanol	Distillation
Example 20	P.E. 9, 8g	1.2g	Ethanol	Distillation

P.E.: Preparation Example

[Examples 21~40] Preparation of polymeric nanoparticles containing coenzyme Q10 using PCL-PEG block copolymer

PCL-PEG block copolymer (Mw = 10,000 dalton, PCL : PEG = 1:1 by weight) and coenzyme Q10 were dissolved in 50ml of organic solvent shown in Table 3, then the mixture solution was poured into 50ml of aqueous solution to induce self-assembling to form nanoparticles. The organic solvent was removed by distillation or dialysis according to the method of Table 3 to obtain aqueous solution of nanoparticles containing coenzyme Q10. The preparation condition of the nanoparticles containing coenzyme Q10 is describe in Table 3.

[Table 3]

	Amphiphilic polymer and amount used	Coenzyme Q10	Organic solvent	Removal of organic solvent
Example 21	P.E. 1, 1.5g	0.5g	Acetone	Distillation
Example 22	P.E. 1, 1.8g	1.2g	Acetone	Distillation
Example 23	P.E. 1, 5.0g	2.5g	Acetone	Distillation
Example 24	P.E. 2, 1.5g	0.5g	Acetone	Distillation
Example 25	P.E. 2, 1.8g	1.2g	Acetone	Distillation
Example 26	P.E. 2, 5.0g	2.5g	Acetone	Distillation
Example 27	P.E. 3, 1.5g	0.5g	Acetone	Distillation
Example 28	P.E. 3, 1.8g	1.2g	Acetone	Distillation
Example 29	P.E. 3, 5.0g	2.5g	Acetone	Distillation
Example 30	P.E. 4, 1.8g	1.2g	Acetone	Distillation
Example 31	P.E. 4, 1.8g	1.2g	Dimethylsulfoxide	Dialysis
Example 32	P.E. 4, 1.8g	1.2g	Dimethylformamide	Dialysis
Example 33	P.E. 4, 1.8g	1.2g	Aceconitryl	Dialysis
Example 34	P.E. 4, 1.8g	1.2g	Tetrahydrofuran	Dialysis
Example 35	P.E. 4, 1.8g	1.2g	Acetic acid	Dialysis
Example 36	P.E. 5, 1.8g	1.2g	Acetone	Distillation
Example 37	P.E. 6, 1.8g	1.2g	Acetone	Distillation
Example 38	P.E. 7, 1.8g	1.2g	Acetone	Distillation
Example 39	P.E. 8, 1.8g	1.2g	Acetone	Distillation
Example 40	P.E. 9, 1.8g	1.2g	Acetone	Distillation

[Example 41~43] Preparation of polymeric nanoparticles containing extract of Thujae occidentalis

Polycaprolactone-co-polyethyleneglycol block copolymer (Mn = 9,200;

polycaprolactone:polyethyleneglycol = 1:1 by weight) and extract of Thujae occidentalis as shown in Table 4 were dissolved in 50g of acetone and stirred homogeneously. After complete dissolution, solution was poured slowly into 50ml of distilled water and stirred. After 1 minute of stirring, then the solution was heated to 50~60℃ and stirred again removing away acetone, and finally obtained dispersion solution of nanoparticles containing the extract of Thujae occidentalis.

[Table 4] Contents of nanoparticles containing the extract of Thujae occidentalis

Component	Example 41	Example 42	Example 43
Polycaprolactone-block-polyethyleneglycol	2.5g	2.5g	2.5g
Extract of Thujae occidentalis	1.5g	2.5g	0.5g

[Example 44] Preparation of polymeric nanoparticles containing minoxidil

2.5g of Poly-D,L-lactic acid-co-polyethyleneglycol block copolymer (Mn = 13,700) and 2.5g of minoxidil were dissolved in solvent mixture consisting of 25g of acetone and 25g ethanol and stirred homogeneously. After complete dissolution, the solution was poured slowly into 50ml of distilled water and stirred. After 1 minute of stirring, the solution was heated to 50~60℃ and stirred again removing away solvent, and finally obtained dispersion solution of nanoparticles containing 2.5g of minoxidil.

[Examples 45~47] Preparation of polymeric nanoparticles containing finasteride, a component for growing and sprouting hairs, by using polycaprolactone-polyethyleneglycol block copolymer

Polycaprolactone-co-polyethyleneglycol block copolymer (Mw = 10,000

dalton, polycaprolactone : polyethyleneglycol = 1:1 by weight) and finasteride were dissolved in 50ml of organic solvent homogeneously. After complete dissolution, the mixture solution was poured into 50ml of aqueous solution to induce self-assembling to form nanoparticles. After 1 minute of stirring, the organic solvent was removed by distillation or dialysis to obtain aqueous solution of nanoparticles containing finasteride. The reaction condition for the preparation of nanoparticles containing finasteride is shown in Table 5.

[Table 5]

	Amphiphilic polymer and amount used (g)	Finasteride	Organic solvent	Removal of organic solvent
Example 45	P.E. 1, 2.5	0.2	Acetone	Distillation
Example 46	P.E. 1, 2.5	1.0	Acetone	Distillation
Example 47	P.E. 1, 2.5	2.0	Acetone	Distillation

[Examples 48-59] Preparation of polymeric nanoparticles containing cyclosporin, a component for growing and sprouting hairs, by using polycaprolactone-polyethyleneglycol block copolymer

Polycaprolactone-co-polyethyleneglycol block copolymer (Mw = 10,000 dalton, polycaprolactone : polyethyleneglycol = 1:1 by weight) and cyclosporin as shown in Table 6 were dissolved in 50ml of organic solvent homogeneously, and the mixture solution was poured into 50ml of aqueous solution to induce self-assembling to form nanoparticles. The organic solvent was removed by distillation or dialysis to obtain aqueous solution of nanoparticles containing cyclosporin.

[Table 6]

	Amphiphilic polymer and amount used (g)	Cyclosporin (g)	Organic solvent	Removal of organic solvent
Example 48	P.E. 1, 1.5	0.5	Acetone	Distillation
Example 49	P.E. 1, 2.0	1.0	Acetone	Distillation
Example 50	P.E. 1, 5.0	0.5	Acetone	Distillation
Example 51	P.E. 2, 1.5	0.5	Acetone	Distillation
Example 52	P.E. 2, 2.0	1.0	Acetone	Distillation
Example 53	P.E. 2, 5.0	0.5	Acetone	Distillation
Example 54	P.E. 3, 1.5	0.5	Ethanol	Distillation
Example 55	P.E. 3, 2.0	1.0	Ethanol	Distillation
Example 56	P.E. 3, 5.0	0.5	Ethanol	Distillation
Example57	P.E. 4, 1.5	0.5	Dimethylsulfoxide	Dialysis
Example58	P.E. 4, 1.8	1.0	Dimethylformamide	Dialysis
Example59	P.E. 4, 1.8	0.5	Acetonitryl	Dialysis

[Experimental Example 1] Measurement of nanoparticle size with dynamic light scattering

Mean sizes of the nanoparticles prepared in Examples 1~59 were measured with Zetasizer 3000Hsa (Malvern, Great Britain). Scattering angle was 90°, and temperature was 25℃. The results are shown in Table 7.

[Table 7]

	Mean size (average diameter; nm)
Example 1	247.2
Example 2	148.2
Example 3	42.2
Example 4	212.4
Example 5	271.8
Example 6	49.3
Example 7	223.4
Example 8	256.8
Example 9	54.3
Example10	243.2
Example11	284.2
Example12	296.1
Example13	276.2
Example14	249.3
Example15	98.9
Example16	95.4
Example17	86.8
Example18	72.3

Example19	53.2
Example20	68.6
Example 21	56.2
Example 22	64.2
Example 23	53.2
Example 24	212.4
Example 25	271.8
Example 26	278.3
Example 27	223.4
Example 28	256.8
Example 29	201.3
Example 30	243.2
Example 31	284.2
Example 32	296.1
Example 33	276.2
Example 34	249.3
Example 35	198.9
Example 36	295.4
Example 37	286.8
Example 38	72.3
Example 39	53.2
Example 40	48.6
Example 41	45.2
Example 42	120.0
Example 43	278.3
Example 44	45.2
Example 45	67.8
Example 46	146.2
Example 47	226.5
Example 48	56.2
Example 49	64.2
Example 50	53.2
Example 51	212.4
Example 52	271.8
Example 53	278.3
Example 54	223.4
Example 55	256.8
Example 56	201.3
Example 57	243.2
Example 58	284.2
Example 59	296.1

[Experimental Example 2] Measurement of skin absorption of nanoparticles containing ginsenoside

Skins of hair Guinea pig were cut and fixed with Franz-diffusion cell, then

the upper position was treated with the 3 kind of samples of Table 8 and the lower position was stayed in a buffer solution while stirring the buffer solution at 32℃ for 18 hours. The amount of Compound K from the ginsenoside absorbed into the skin was measured with liquid chromatography.

[Table 8]

	Amount of ginsenoside absorbed	Area of ginsenoside and absorption amount per hour
Ethanol(10%), Butyleneglycol (5%), Ginsenoside (1%)	366㎍	40㎍/㎠ · h
Example 3	576㎍	64㎍/㎠ · h
Microemulsion containing 1% ginsenoside	485㎍	54㎍/㎠ · h

As can be seen in the above results, the nanoparticle prepared in Example 3 of the present invention has better absorption property than microemulsion formulation about 159%.

[Experimental Example 3] Measurement of skin absorption of nanoparticles containing coenzyme Q10

Skins of hair Guinea pig were cut and fixed with Franz-diffusion cell, then the upper position was treated with the 3 kind of samples of Table 9 and the lower position was stayed in a buffer solution while stirring the buffer solution at 32℃ for 18 hours. The amount of coenzyme Q10 absorbed into the skin was measured with liquid chromatography. In order for comparison, same experiment was performed for the liposome containing 1% of coenzyme Q10.

[Table 9]

	Amount of coenzyme Q10 absorbed	Area of coenzyme Q10 and absorption amount per hour
10% solution of capric/caprylic triglyceride	0	0
Example 24	37.53㎍	2.68㎍/㎠ · h
Liposome containing 1% of coenzyme Q10	23.68㎍	1.69㎍/㎠ · h

As can be seen in Table 9, the nanoparticle containing coenzyme Q10 of the present invention has better absorption property than liposome about 159%.

[Experimental Example 4] Measurement of skin absorption of nanoparticles containing minoxidil

In the procedure preparing the nanoparticles of Example 44, a fluorescent material, Rubren, was added as a probe (label) and the nanoparticles prepared were applied on the skin of haircut hairy Guinea pig and on the flank organ of a hamster with a closed patch for 6 hours. Obtained tissues were cut with 40㎛ thickness to prepare cryosection, then dyed with DAPI to mark nucleated cells (karyota), and the amount of Rubren absorbed into the skin through hair bulb was measured with confocal laser scanning microscopy (Zeiss).

From the above result, it was found that the concentration gradient of the nanoparticles containing minoxidil absorbed into the skin was same throughout the skin, the concentration was high near the hair bulb and the nanoparticles were absorbed through hair bulb.

[Experimental Example 5] Measurement of skin absorption of nanoparticles containing cyclosporin

Skins of hair Guinea pig were cut and fixed with Franz-diffusion cell, then the upper position was treated with the 3 kind of samples of Table 10 and the lower position was stayed in a buffer solution while stirring the buffer solution at 32℃ for 18 hours. The amount of cyclosporin absorbed into the skin was measured with liquid chromatography.

[Table 10]

	Amount of cyclosporin absorbed	Area of cyclosporin and absorption amount per hour
10% solution of capric/caprylic triglyceride	0	0
Example 51	37.53μg	2.68μg/㎠ · h
Liposome[1] containing 1% of cyclosporin	23.68μg	1.69μg/㎠ · h

[Note] Liposome[1] is a comparative experimental sample.

As can be seen in Table 10, the nanoparticle of Example 51 of the present invention has better absorption property than liposome.

[Experimental Example 6] Effects of nanoparticles containing minoxidil or extracts of Thujae occidentalis in the growth of hairs

In order to test hair-growing effects, the effects of the nanoparticles prepared in the present Examples were compared with those of hair-growing components not captured in the nanoparticles.

Hairs on the backs of the mice 47~53 days from birth (C57BL/6) were removed, and 100μℓ of the test samples were applied on the backs of the mice, 10 mice per sample, everyday.

Hair-growing effects were valuated according to the length of hairs and

degree of growth after removal of hairs scoring 0 to 3. In or to compare the hair-growing effects, 30% alcohol solution was applied to each mouth as a control. The results are shown in Table 11.

[Table 11]

Test material	Time	
	After 10 days	After 20 days
Negative control (ethanol)	0.25±0.25	1.42±0.30
Positive control (minoxidil 2.5%)	1.22±0.24	2.65±0.47
Extract of Thujae occidentalis (2.5%)	0.67±0.41	1.98±0.52
Nanoparticles of Example 41	0.91±0.28	2.56±0.32
Nanoparticles of Example 42	0.82±0.32	2.42±0.23
Nanoparticles of Example 43	0.76±0.15	2.13±0.40
Nanoparticles of Example 44	1.65±0.43	2.86±0.42

As can be seen in the above result, the nanoparticle of the Example 44 of the present invention containing minoxidil showed better hair-growing effect than the minoxidil of the same concentration, in addition, the nanoparticle of the Examples 41~43 of the present invention containing extracts of Thujae occidentalis showed better hair-growing effect than the extracts of Thujae occidentalis. From the results of Examples 41~43, it was found that the concentration of the active components and the ratio of the nanoparticles are important for the sprout or growth of the hairs.

[Experimental Example 7] Effects of nanoparticles containing finasteride in the growth of hairs

In order to test hair-growing effects, the effects of the nanoparticles prepared in the present Examples 45~47 were compared with those of hair-growing components not captured in the nanoparticles.

Hairs on the backs of the mice 47~53 days from birth (C57BL/6) were removed, and 100㎕ of the test samples were applied on the backs of the mice, 10 mice per sample, everyday.

Hair-growing effects were valuated according to the length of hairs and degree of growth after removal of hairs scoring 0 to 3. In or to compare the hair-growing effects, same amount (1%) of finasteride was dissolved in 30% alcohol solution and applied to each mouth as a negative control and, cyclosporin was dissolved therein as a positive control. The results are shown in Table 12.

[Table 12]

Test material	Time	
	After 10 days	After 20 days
Negative control	0.26±0.24	1.52±0.28
Positive control (0.5% of cyclosporin)	1.05±0.35	2.55±0.27
Nanoparticles of Example 46	0.55±0.23	1.95±0.22

As can be seen in the above result, even though the nanoparticle of the present Example containing finasteride showed lower hair-growing effect than positive control containing 5% of cyclosporin, but showed better hair-growing effect than that of the finasteride dissolved in ethanol with a same concentration.

[Experimental Example 8] Effects of nanoparticles containing cyclosporin in the growth of hairs

In order to test hair-growing effects, the effects of the nanoparticles prepared in the Examples 48~59 were compared with those of hair-growing components not captured in the nanoparticles.

Hairs on the backs of the mice 47~53 days from birth (C57BL/6) were

removed, and 100㎕ of the test samples were applied on the backs of the mice, 10 mice per sample, everyday.

Hair-growing effects were valuated according to the length of hairs and degree of growth after removal of hairs scoring 0 to 3. In or to compare the hair-growing effects, 30% alcohol solution was applied to each mouth as a control. The results are shown in Table 13.

[Table 13]

Test material	Time	
	After 10 days	After 20 days
Negative control (ethanol)	0.26± 0.24	1.52± 0.28
Positive control (0.5% of cyclosporin)	1.05± 0.35	2.55± 0.27
Nanoparticles of Example 48	1.18± 0.22	2.68± 0.32
Nanoparticles of Example 51	1.24± 0.26	2.82± 0.21
Nanoparticles of Example 54	1.34± 0.15	2.86± 0.16

As can be seen in the above result, the nanoparticle of the Examples of the present invention showed better hair-growing effect than effective component of hair-growing itself at the same concentration, in addition, from the results of Examples 48, 51 and 54, it was found that the concentration of the active components are important in the sprout or growth of the hairs.

[Experimental Example 9] Anti-oxidation effect of nanoparticles containing coenzyme Q10 to the skin cell

100㎕ of HCSS (HEPES-buffered control salt solution) was applied to the fibroblast of the dermis according to the concentration of Table 14, and the fluorescence of the dichlorofluorescein (DCF) initially oxidated to ROS (active oxygen species) was measured with fluorescent plate reader (Ex=485nm,

Em=530nm). UVB (30mJ/㎠) was irradiated thereto, and the fluorescence was measured with fluorescent plate reader (Ex=485nm, Em=530nm) immediately after irradiation and after 3 hours from irradiation. In order for comparison, same experiment was performed to the liposome containing 1% of coenzyme Q10. Table 14 shows the result of comparison (%) of fluorescence with that of control not treated with sample after 3 hours from UVB irradiation by % unit.

[Table 14]

Samples	Concentration				
	10 ppm	5 ppm	2.5 ppm	1.25 ppm	0.625 ppm
Example 24	62.6	67.6	74.8	79.1	74.9
Liposone containing 1% of coenzyme Q10	87.1	93.8	96.3	89.4	85.5

As can be seen in Table 14, suppression of generation of ROS is more effective in the nanoparticle containing coenzyme Q10 of the present invention than in the liposome containing coenzyme Q10.

[Experimental Example 10] Biosynthesis of collagen by the nanoparticles containing ginsenoside

Human fibroblast was cultured in 24 well plate culture, and nanoparticles prepared by the method of Example 3 and microemulsions prepared according to following Comparative Example 1, which containing ginsenosides of the Table 15, were diluted to 1/100 and added to the culture. After 3 days of culture, 0.5ml of DMEM (Dulbecos modified eagles medium) containing 10% of fetal bovine serum (FBS) was added, and 10μg· Ci of L[2,3,4,5-3H]-proline was added.

After 24 hours later, cells and medium of each well were gathered and washed in 5% of TCA (Trichloroacetic acid), then separated into 2 test tubes; 1unit/㎕ of type I collagenase was added to one test tube and cultured for 90 minutes at 37℃, and the other test tube was stayed at 4℃.

Then, 0.05ml of 50% TCA was added all of the test tubes and stayed for 20 minutes at 4℃, and centrifuged at 12000rpm for 10 minutes, then DPM (Decay per minute) values of the supernatant and the precipitate were measured with scintillation counter, and biosynthesis of collagen by the formulations of the Example 3 and the Comparative Example 1 containing same amount of ginsenoside were calculated according to Calculation Formula 1. The results are in Table 15

[Calculation Formula 1]

RCB={collagen DMP / (total collagen DMP – collagen DMP) x 5.4 + collagen DMP} x 100

[Comparative Example 1]

Component	Comparative Example 1
Hydrated lecithin	2.5
Hydrated lysophosphatidylcholine	0.15
Propylene glycol	4.0
Ethanol	6.5
Ginsenoside	1.5
EDTA	0.05
Glycerin	4.0
Betaine	1.0
Distilled water	to 100

[Table 15]

Concentration of ginsenoside (%)	Increase of collagen biosynthesis (%)	
	Example 3	Comparative Example 1
$1x\ 10^{-8}$	7	5
$1x\ 10^{-7}$	35	24
$1x\ 10^{-6}$	40	35
$1x\ 10^{-5}$	59	45
$1x\ 10^{-4}$	73	50

As can be seen in Table 15, ginsenoside captured in the nanoparticle of the present invention shows more excellent property of promoting biosynthesis of collagen compared with that of the ginsenoside not captured.

Following formulations were prepared by using the above Examples.

[Formulations 1~9] Cream Formulation

O/W Emulsion formulations comprising nanoparticles of the Examples containing ginsenoside are shown in Table 16.

[Table 16]

Component	Form. 1	Form. 2	Form. 3	Form. 4	Form. 5	Form. 6	Form. 7	Form. 8	Form. 9
Lipophilic monostearic acid	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Glyceryl stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Cetostearyl alcohol	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Paraffin	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Squalene	1.2	1.2	1.2	1.2	1.2	1.2	1.2	1.2	1.2
Polysorbate 60	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Cetylethylhexanoate	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Liquid paraffin	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Mehtyl paraben	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Propyl paraben	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Silicon oil	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
Tocopheryl acetate	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Distilled water	To 100	To 100	To 100	To 100	To 100	To 100	To 100	To 100	To 100
Urea	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Glycerin	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0

Butyleneglycol	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Triethanolamine	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Carboxyvinyl polymer	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Example 1	25.0	-	-	-	-	-	-	-	-
Example 4	-	25.0	-	-	-	-	-	-	-
Example 5	-	-	25.0	-	-	-	-	-	-
Example 6	-	-	-	25.0	-	-	-	-	-
Example 8	-	-	-	-	25.0	-	-	-	-
Example 10	-	-	-	-	-	25.0	-	-	-
Example 16	-	-	-	-	-	-	25.0	-	-
Example 17	-	-	-	-	-	-	-	25.0	-
Example 18	-	-	-	-	-	-	-	-	25.0

Form.: Formulation

[Formulation 10~18] Soft water (Skin softener) Formulation

Soft water formulations comprising nanoparticles of the Examples containing ginsenoside are shown in Table 17.

[Table 17]

Component	Form. 10	Form. 11	Form. 12	Form. 13	Form. 14	Form. 15	Form. 16	Form. 17	Form. 18
Betaine	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Natto gum	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Cellulose gum	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
Ethanol	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Polyoxyethylene Hydrogenated castor oil	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Tocopherylacetate	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Polysorate 60	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Glycerin	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Example 1	10.0	-	-	-	-	-	-	-	-
Example 2	-	10.0	-	-	-	-	-	-	-
Example 3	-	-	10.0	-	-	-	-	-	-
Example 4	-	-	-	10.0	-	-	-	-	-
Example 5	-	-	-	-	10.0	-	-	-	-
Example 6	-	-	-	-	-	10.0	-	-	-
Example 15	-	-	-	-	-	-	10.0	-	-
Example 19	-	-	-	-	-	-	-	10.0	-
Example 20	-	-	-	-	-	-	-	-	10.0
Preservative	small	small	small	small	small	small	small	small	small
Pigment	small	small	small	small	small	small	small	small	small
Distilled water	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100

[Formulation 19~27] Cream Formulation

O/W Emulsion formulations comprising nanoparticles of the Examples 21, 24~26, 28, 30, 36~38 containing coenzyme Q10 are shown in Table 18.

[Table 18]

Component	Formulation								
	19	20	21	22	23	24	25	26	27
Lipophilic monostearic acid	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Glyceryl stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Cetostearyl alcohol	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Paraffin	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Squalene	1.2	1.2	1.2	1.2	1.2	1.2	1.2	1.2	1.2
Polysorbate 60	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Cetylethylhexanoate	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Liquid paraffin	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Mehtyl paraben	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Propyl paraben	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Silicon oil	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
Tocopheryl acetate	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Distilled water	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100
Urea	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Glycerin	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Butyleneglycol	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Triethanolamine	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Carboxyvinyl polymer	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Example 21	25.0	-	-	-	-	-	-	-	-
Example 24	-	25.0	-	-	-	-	-	-	-
Example 25	-	-	25.0	-	-	-	-	-	-
Example 26	-	-	-	25.0	-	-	-	-	-
Example 28	-	-	-	-	25.0	-	-	-	-
Example 30	-	-	-	-	-	25.0	-	-	-
Example 36	-	-	-	-	-	-	25.0	-	-
Example 37	-	-	-	-	-	-	-	25.0	-
Example 38	-	-	-	-	-	-	-	-	25.0

[Formulation 28~36] Skin Formulation

Skin formulations comprising polymeric nanoparticles of the Examples 21~26, 35, 39, 40 are shown in Table 19.

[Table 19]

Components	Formulation								
	28	29	30	31	32	33	34	35	36
Betaine	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Natto gum	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Cellulose gum	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
Ethanol	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Polyoxyethylene hydrogenated castor oil	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Tocopherylacetate	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Polysorate 60	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Glycerin	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Example 21	10.0	-	-	-	-	-	-	-	-
Example 22	-	10.0	-	-	-	-	-	-	-
Example 23	-	-	10.0	-	-	-	-	-	-
Example 24	-	-	-	10.0	-	-	-	-	-
Example 25	-	-	-	-	10.0	-	-	-	-
Example 26	-	-	-	-	-	10.0	-	-	-
Example 35	-	-	-	-	-	-	10.0	-	-
Example 39	-	-	-	-	-	-	-	10.0	-
Example 40	-	-	-	-	-	-	-	-	10.0
Preservative	small	small	small	small	small	small	small	small	small
Pigment	small	small	small	small	small	small	small	small	small
Distilled water	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100	to 100

[Experimental Example 11] Storage of coenzyme Q10 in the formulation

Stability of of coenzyme Q10 in the formulation during long terms of storage verified with Formulation 20. Storing the formulations in thermostatic baths of 25℃ and 45℃, samples were taken and the quantities of coenzyme Q10 were measured. In order for comparison, cream formulation comprising liposome containing 1% of coenzyme Q10 was prepared with reference to the composition of Table 18, and same test was performed. The results are shown in Table 20.

[Table 20]

	Condition	7 days	15 days	30 days	45 days
Formulatio 20	25℃	100%	94%	90%	87%
	45℃	100%	91%	86%	83%
Cream formulation comprising liposome	25℃	100%	93%	88%	82%
	45℃	100%	87%	75%	62%

As can be seen in Table 20, coenzyme Q10 in the polymeric nanoparticle of the present invention is more stable than that in the liposome during long terms of storage. Therefore, it is possible to maintain the activity of coenzyme Q10 by capturing coenzyme Q10 in the polymeric nanoparticle of the present invention.

[Formulation 37] Skin Formulation

Skin formulation comprising nanoparticles of the Example 44 containing minoxidil is shown in Table 21.

[Table 21]

Component	Contents (Wt%)
Water	To 100
EDTA-2Na	0.02
DL-panthenol	0.1
Trehalose	2.0
Glycerin	3.0
Butylent glycol	3.0
PEG-1500	1.0
Rose water	1.0
Bio-HE	0.01
Soypol sp	0.01
Ethanol	5.0
Propyl paraben	0.02
Methyl paraben	0.12
Tagat CH 60	0.35
C-940	6.0
SPA base	0.5
KOH (10%)	0.24
Nanoparticle of Example 4	25.0

[Formulation 38] Hair tonic Formulation

Hair tonic composition comprising nanoparticles of the Examples 41~43 containing extracts of Thujae occidentalis and nanoparticles of Example 44 containing minoxidil is shown in Table 22.

[Table 22]

Component	Contents (Wt%)
Ethanol	55.0
Caster oil	5.0
Glycerin	3.0
Piroctone Olamine	0.1
Nanoparticles of Examples 41~44	1.0
Perfume, Pigment	Suitable
Distilled water	to 100

[Formulation 39] Hair liquid Formulation

Hair liquid composition comprising nanoparticles of the Examples 41~43 containing extracts of Thujae occidentalis and nanoparticles of Example 44 containing minoxidil is shown in Table 23.

[표 23]

Component	Contents (Wt%)
Polyoxypropylbutylether (40 P.O.)	15.0
Polyoxypropylbutylether phosphate (40 P.O.)	15.0
1,3-butylene glycol	5
95% ethanol	50
Tocopheryl acetate	0.1
Nanoparticles of Examples 41~43	5.0
Perfume,	Suitable
Pigment	Suitable
Edetic acid	Suitable
Distilled water	to 100

[Formulation 40] Scalp treatment Formulation

Scalp treatment composition comprising nanoparticles of the Examples 41~43 containing extracts of Thujae occidentalis and nanoparticles of Example 44 containing minoxidil is shown in Table 24.

[Table 24]

Component	Contents (wt%)
1,3-butylene glycol	0.5
Tetra-2-ethylhexapentaerythritol	1.2
95% ethanol	60
Nanoparticles of Examples 41~44	2
Perfume	Suitable
Dimethylether/LPG(95/5)	to 100

[Formulation 41] Hair cream Formulation

Hair cream composition comprising nanoparticles of the Examples 41~44 is shown in Table 25.

[Table 25]

Component	Contents (wt%)
Liquid paraffin	5.0
Cetylstearyl alcohol	5.5
Vaseline	5.5
Glycerylmonostearate	3.0
Polyoxyethylene(20mole added)-2-octyldodecylether	3.0
Tocopheryl acetate	0.05
Propylparaben	0.3
Nanoparticles of Examples 41~44	5
Polyethyleneglycol 4000	5.0
Glycerin	7.0
Perfume	Suitable
Distilled water	to 100

[Formulation 42] Hair gel Formulation

Hair gel composition comprising nanoparticles of the Examples 41~44 is shown in Table 26.

[Table 26]

Component	Contents (wt%)
Carboxyvinyl polymer	0.7
polyvinylpyrolidone	2.0
Glycerin	4.0
Sodium hydroxide	Suitable
Ethanol	20.0
Nanoparticles of Examples 41~44	2
polyoxyethyleneoctyldodecylether	Suitable
Perfume	Suitable
Edetic acid	Suitable
Distilled water	to 100

[Formulation 43] Hair spray Formulation

Hair spray composition comprising nanoparticles of the Examples 41~44 is shown in Table 27.

[Table 27]

Component	Contents (wt%)
Acryl resin alkanolamine salt (50%)	3.5
Cetylalcohol	0.05
Silicon oil (methylphenylpolysiloxane)	0.15
Ethanol	45
Nanoparticles of Examples 41~44	1
Perfume	Suitable
Dimethylether	45
LPG	5.0

[Formulation 44] Hair shampoo Formulation

Hair shampoo composition comprising nanoparticles of the Examples 41~44 is shown in Table 28.

[Table 28]

Component	Contents (wt%)
Sodiumlaurate sulfate	16
Cocoaminopropyl betaine	2
Jaguar C13S	0.1
Nanoparticles of Examples 41~44	3
Distilled water	to 100

[Formulation 45] Cream Formulation

O/W emulsion composition comprising nanoparticles of the Example 46 is shown in Table 29.

[Table 29]

Component	Contents (wt%)
Liquid paraffin	5.0
Cetylstearyl alcohol	5.5
Vaseline	5.5
Glycerylmonostearate	3.0
Polyoxyethylene(20mole added)-2-octyldodecylether	3.0
Tocopheryl acetate	0.05
Propylparaben	0.3
Polyethyleneglycol 4000	5.0
Glycerin	7.0
Perfume	Suitable
Distilled water	to 100
Example 46	1

[Formulation 46] Tonic Formulation

O/W emulsion composition comprising nanoparticles of the Example 46 is shown in Table 30.

[Table 30]

Component	Contents (wt%)
Ethanol	55.0
Castor oil	5.0
Glycerin	3.0
Pyroctoneamine	0.1
Perfume, Pigment	Suitable
Distilled water	to 100
Example 46	0.5

[Formulations 47~52] Cream Formulation

O/W emulsion composition comprising nanoparticles of the Examples 48, 51~53, 55, 57 is shown in Table 31.

[Table 31]

Component	Form. 47	Form. 48	Form. 49	Form. 50	Form. 51	Form. 52
Liquid paraffin	5.0	5.0	5.0	5.0	5.0	5.0
Cetylstearyl alcohol	5.5	5.5	5.5	5.5	5.5	5.5
Vaseline	5.5	5.5	5.5	5.5	5.5	5.5
Glycerylmonostearate	3.0	3.0	3.0	3.0	3.0	3.0
Polyoxyethylene (20mole added)-2-octyldodecylether	3.0	3.0	3.0	3.0	3.0	3.0
Tocopheryl acetate	0.05	0.05	0.05	0.05	0.05	0.05
Propylparaben	0.3	0.3	0.3	0.3	0.3	0.3
Polyethyleneglycol 4000	5.0	5.0	5.0	5.0	5.0	5.0
Glycerin	7.0	7.0	7.0	7.0	7.0	7.0
Perfume	Suitable	Suitable	Suitable	Suitable	Suitable	Suitable
Distilled water	to 100	to 100	to 100	to 100	to 100	to 100
Example 48	5					
Example 51		5				
Example 52			5			
Example 53				5		
Example 55					5	
Example 57						5

Form.: Formulation

[Formulations 53~58] Tonic Formulation

O/W emulsion composition comprising nanoparticles of the Examples 1, 4~6, 8, 10 is shown in Table 32.

[표 32]

Component	Form. 53	Form. 54	Form. 55	Form. 56	Form. 57	Form. 58
Ethanol	55.0	55.0	55.0	55.0	55.0	55.0
Castor oil	5.0	5.0	5.0	5.0	5.0	5.0
Glycerin	3.0	3.0	3.0	3.0	3.0	3.0
Pyroctoneamine	0.1	0.1	0.1	0.1	0.1	0.1
Perfume, Pigment	Suitable	Suitable	Suitable	Suitable	Suitable	Suitable
Distilled water	to 100	to 100	to 100	to 100	to 100	to 100
Example 1	2					
Example 4		2				
Example 5			2			
Example 6				2		
Example 8					2	
Example 10						2

CLAIMS

1. A self-assembled polymeric nanoparticle comprising amphiphilic polymer and physiologically active ingredient, wherein the amphiphilic polymer comprises polycaprolactone as hydrophobic block and polyethyleneglycol as hydrophilic block and entraps the physiologically active ingredient.

2. The self-assembled polymeric nanoparticle according to claim 1, wherein the size of the nanoparticle is 1~1,000nm.

3. The self-assembled polymeric nanoparticle according to claim 1, wherein the amount of the physiologically active ingredient is 0.1~50wt% to the total weight of the nanoparticle.

4. The self-assembled polymeric nanoparticle according to claim 1 wherein the amphiphilic polymer is copolymer of polycaprolactone and polyethyleneglycol with a ratio of 1:9 to 9:1 by weight.

5. The self-assembled polymeric nanoparticle according to claim 1, wherein molecular weight of the polycaprolactone is 500 to 100,000 daltons.

6. The self-assembled polymeric nanoparticle according to claim 1, wherein molecular weight of the polyethyleneglycol is 500 to 100,000 daltons.

7. The self-assembled polymeric nanoparticle according to claim 1, wherein the physiologically active ingredient is selected from the group consisting of *Rheum undulatum*, genistein, hesperetin, hesperidine, catechin, isoflavone, danazol, haloperidol, furosemid, isosorbide dinitrate, chloramfenicol, sulfamethoxazole, caffeine, cimethidine, diclofenac Na, coenzyme Q10, vitamin E and its derivatives, vitamin A and its derivatives, provitamin D_3 and its derivatives, ursolic acid, oleanolic acid, rosmarinic acid, 18 beta-glycyrrhetinic acid, glabridin, aleuritic acid, polyphenol, esculin, (-) epigallocatechin gallate, turmeric acid, ginsenosides, tetra hydrocurcuminoids, centella asiatica, beta carotene, asiaticoside, farnesol, beta-sitosterol, linoleic acid, gamma linolenic acid, resveratrol, vineatrol, ginkgo biloba, triclosan, minoxidil, natural oil, ceramide, sphingosine, extracts of Thujae occidentalis, extracts of Polygoni multiflori Radix, extracts of Glycyrrhiza uralensis, extracts of Coix lachryma-jobi var. ma-yuen and finasteride.

8. The self-assembled polymeric nanoparticle according to claim 1, wherein the physiologically active ingredient is insoluble in water.

9. The self-assembled polymeric nanoparticle according to claim 8, wherein the physiologically active ingredient is selected from the group consisting of ginsenoside, coenzyme Q10 and hair growing or sprouting component.

10. The self-assembled polymeric nanoparticle according to claim 9, wherein

the physiologically active ingredient is selected from the group consisting of finasteride, minoxidil, extracts of Thujae occidentalis, extracts of polygoni multiflori Radix, extracts of Glycyrrhiza uralensis, extracts of Coix lachryma-jobi var. ma-yuen, isoflavone, genistein, hesperetin, hesperidine, catechin, vitamin E and its derivatives, vitamin A its derivatives, provitamin D3 and its derivatives, ursolic acid, oleanolic acid, rosmarinic acid, 18-beta-glycyrrhetinic acid, farnesol, beta- sitosterol, linoleic acid, gamma linolenic acid, resveratrol, ceramide, Sphingosine and ginsenoside.

11. A skin external composition comprising the nanoparticle according to any one of claims 1 to 10.

12. The skin external composition according to claim 11, which has a formulation of hair tonic, hair liquid, scalp treatment, hair cream, hair gel, hair spray, hair shampoo, rinse, soft water, skin softener, nutrition water, nutrition cream, massage cream, essence, eye cream, eye essence, cleansing cream, cleansing foam, cleansing water, pack, powder, make-up base, foundation, body lotion, body cream, body oil, body essence, hair dye, lotion, ointment, gel, cream, patch or spray.

13. A method for preparing self-assembled polymeric nanoparticle comprising the steps of:

Preparing amphiphilic polymer by co-polymerizing polycaprolactone as a hydrophobic block and polyethyleneglycol as a hydrophilic;

Dissolving the above amphiphilic polymer and a physiologically active component in an organic solvent and stirring to prepare solution mixture;

Adding the solution mixture prepared into an aqueous solution to emulsify; and

Removing organic solvent to obtain nanometer-sized particle.

14. The method according to claim 13, wherein the amphiphilic polymer is copolymer of polycaprolactone and polyethyleneglycol with a ratio of 1:9 to 9:1 by weight.

15. The method according to claim 13, wherein the physiologically active component is insoluble to water.

16. The method according to claim 13, wherein the physiologically active component is selected from the group consisting of ginsenoside, coenzyme Q10 and hair growing or sprouting component.

FIGURES

FIG. 1

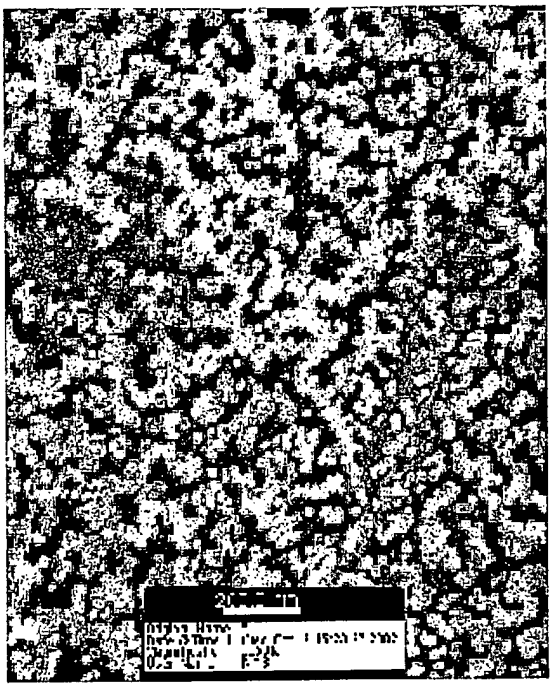


FIG. 2

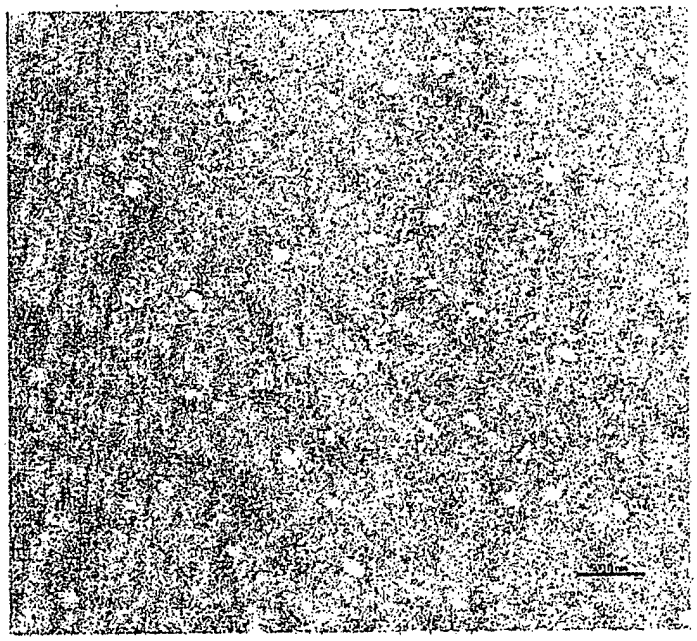


FIG. 3

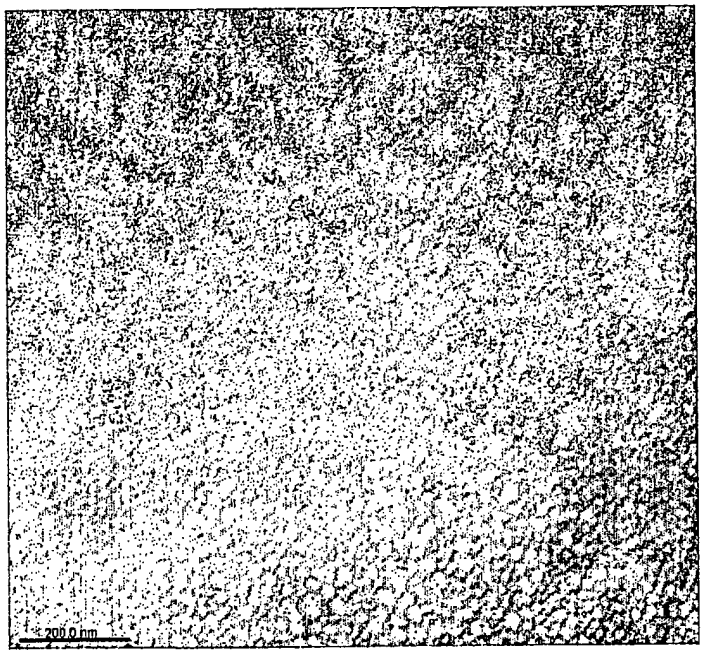


FIG. 4

FIG. 5

INTERNATIONAL SEARCH REPORT

International application No.
PCT/KR2004/001572

A. CLASSIFICATION OF SUBJECT MATTER

IPC7 A61K 7/06

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
ICP7 A61K 7/06, A61K 9/127, A61K 9/50, A61K 9/16, A61K 31/74, A61K 31/70

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
Korean Patents and applications for inventions since 1975

Electronic data base consulted during the intertnational search (name of data base and, where practicable, search terms used)
STN, PUBMED, DELPHION, KISTI

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	KR 1999-69033 A (NIHON-MEDI PHYSICS CO., LTD.) 06 SEPTEMBER 1999 see the whole document	1-16
Y	WO 02/45689 A1 (SAMYANG CO., LTD.) 13 JUNE 2002 see the whole document	1-16
Y	US 5543158 (MASSACHUSETTS INSTITUTE OF TECHNOLOGY) 06 AUGUST 1996 see the whole document	1-16
Y	KR 2003-45611 A (SAMYANG COP.) 11 JUNE 2003 see the whole document	1-16
A	US 4474753 (MERCK & CO., INC.) 02 OCTOBER 1984 see the whole document	1-16

☐ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" earlier application or patent but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents,such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
22 OCTOBER 2004 (22.10.2004)	**25 OCTOBER 2004 (25.10.2004)**
Name and mailing address of the ISA/KR Korean Intellectual Property Office 920 Dunsan-dong, Seo-gu, Daejeon 302-701, Republic of Korea Facsimile No. 82-42-472-7140	Authorized officer JUNG, YOUNG JA Telephone No. 82-42-481-8164

Form PCT/ISA/210 (second sheet) (January 2004)

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No.
PCT/KR2004/001572

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
KR 1999-69033 A	06.09.1999	NONE	
WO 0245689 A1	13.06.2002	US 20040037885 A1	2004-02-26
		KR 2045569 A	2002-06-19
		JP 2004514734 T2	2004-05-20
		EP 1339389 A1	2003-09-03
		CN 1477952 T	2004-02-25
		CA 2430481 AA	2002-06-13
		AU 0222733 A5	2002-06-18
US 5543158	06.08 1996	WO 9503357 A1	1995-02-02
		WO 9503356 A1	1995-02-02
		US 6007845	1999-12-28
		US 5578325	1996-11-26
		US 5565215	1996-10-15
		US 5543158	1996-08-06
		JP 09504308 T2	1997-04-28
		JP 09504042 T2	1997-04-22
		EP 0712421 A1	1996-05-22
		EP 0710261 B1	2004-05-12
		EP 0710261 A1	1996-05-08
		DE 69433775 C0	2004-06-17
		CA 2167921 AA	1995-02-02
		CA 2167920 AA	1995-02-02
		AT 0266695 E	2004-05-15
KR 2003-45611 A	11.06.2003	NONE	
US 4474753	02.10.1984	US 4478822	1984-10-23
		JP 59219217 A2	1984-12-10
		GB 8316437 A0	1983-07-20
		ES 8601706 A1	1986-03-01
		ES 0532317 A1	1985-11-16
		EP 0126684 B1	1991-08-28
		DK 0233884 A0	1984-05-07
		DE 3484969 C0	1991-10-02
		CA 1224148 A1	1987-07-14
		AU 1937883 A1	1984-11-08
		AT 0066617 E	1991-09-15